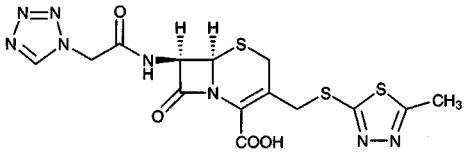
Cefazolin



Molecular formula: $C_{14}H_{14}N_8O_4S_3$
Molecular weight: 454.51
CAS Registry No.: 25953-19-9, 27164-46-1 (sodium salt)
Merck Index: 1967
Lednicer No.: 3 442

SAMPLE

Matrix: blood
Sample preparation: Condition an 800 µL 500 mg Sep-Pak Vac 3cc C18 SPE cartridge twice with 800 µL MeOH and with 800 µL water. 200 µL Serum + 200 µL water, vortex. Add to the SPE cartridge. Wash twice with 800 µL water. Elute twice with 400 µL MeCN: 50 mM KH_2PO_4 70:30 and twice with 400 µL MeCN:water 50:50. Evaporate the eluate under a stream of nitrogen at 50° for 10 min. Cool, centrifuge at 1450-1475 g for 4 min, inject a 50 µL aliquot of the supernatant.

HPLC VARIABLES

Guard column: KGCQ-324C (YMC, Wilmington, NC)
Column: 250 × 4.6 5 µm YMC pack ODS-AQ (YMC, Wilmington, NC)
Mobile phase: Gradient. A was MeCN:MeOH:50 mM pH 6 phosphate buffer 5:4:91. B was MeCN:MeOH:50 mM pH 6 phosphate buffer 8:8:84. A:B 100:0 for 2 min, to 0:100 over 9 min, maintain at 0:100 for 14 min, to 100:0 over 5 min, maintain at 100:0 for 5 min (Prepare buffer as follows. Dissolve 11.94 g KH_2PO_4 and 2.14 g K_2HPO_4 in 2 L water.)
Flow rate: 1.5
Injection volume: 50
Detector: UV 210

CHROMATOGRAM

Retention time: 27
Internal standard: cefazolin

OTHER SUBSTANCES

Extracted: vancomycin
Simultaneous: acetaminophen, salicylates, theophylline

KEY WORDS

serum; SPE; cefazolin is IS

REFERENCE

Backes,D.W.; Aboleneen,H.I.; Simpson,J.A. Quantitation of vancomycin and its crystalline degradation product (CDP-1) in human serum by high performance liquid chromatography, *J.Pharm.Biomed.Anal.*, **1998**, *16*, 1281–1287.

SAMPLE

Matrix: blood
Sample preparation: Mix 100 µL plasma with 40 µL 5% trichloroacetic acid in MeOH. Vortex for 10 s, add 50 µL 30 µg/mL IS. Vortex for 10 s, centrifuge at 5000 rpm for 10 min, inject an aliquot.

HPLC VARIABLES

Column: 300 × 3.9 µBondapak C18
Mobile phase: MeCN:50 mM KH_2PO_4 7:93
Flow rate: 3
Detector: UV 273

CHROMATOGRAM
Retention time: 14
Internal standard: cefoxitin (11)
Limit of detection: 500 ng/mL
Limit of quantitation: 2 μg/mL

KEY WORDS
plasma; pharmacokinetics; rat

REFERENCE
Allababidi,S.; Shah,J.C. Efficacy and pharmacokinetics of site-specific cefazolin delivery using biodegradable implants in the prevention of post-operative wound infections, *Pharm.Res.*, **1998**, *15*, 325–333.

SAMPLE
Matrix: blood
Sample preparation: Dilute serum with an equal volume of water, inject a 20 μL aliquot onto column A, elute column A to waste with MeOH:10 mM pH 7.0 phosphate buffer 5:95 at 0.3 mL/min, after 1.3 min elute the contents of column A onto column B with mobile phase A or B, elute with mobile phase A or B, monitor the effluent from column B.

HPLC VARIABLES
Column: A 50 × 2.1 40 μm Supelclean LC-NH_2; B 150 × 4.6 3 μm Supelcosil LC-18
Mobile phase: A MeCN:MeOH:10 mM pH 7.0 phosphate buffer 15:20:65 containing 5 mM tetrabutylammonium hydrogen sulfate; B MeOH:10 mM pH 7.0 phosphate buffer 30:70 containing 5 mM tetrabutylammonium hydrogen sulfate
Flow rate: 1
Injection volume: 20
Detector: UV 267

CHROMATOGRAM
Retention time: 5.0 (mobile phase A), 6.0 (mobile phase B)
Limit of detection: 500-2000 ng/mL

OTHER SUBSTANCES
Extracted: cefamandole, cefodizime, cefoperazone, cefoxitin, ceftizoxime, ceftriaxone, cefuroxime, cephaloridine, cephalothin
Noninterfering: acetaminophen, acyclovir, digoxin, fluconazole, ranitidine, teicoplanin, theophylline, vancomycin

KEY WORDS
column-switching; serum

REFERENCE
Bompadre,S.; Ferrante,L.; Leone,L. On-line solid-phase extraction of cephalosporins, *J.Chromatogr.A*, **1998**, *812*, 191–196.

SAMPLE
Matrix: blood
Sample preparation: Mix serum with an equal volume of 250 μg/mL 4'-nitroacetanilide in MeCN:MeOH 90:10, mix, let stand at room temperature for 10 min, mix, centrifuge at 12800 *g* for 2 min, inject a 25 μL aliquot of the supernatant.

HPLC VARIABLES
Guard column: RCSS Guard-Pak (Waters)
Column: 100 × 8 C18 Radial Pak (Waters)
Mobile phase: MeOH:0.75% acetic acid 30:70, pH adjusted to 5.5 with triethylamine

Flow rate: 3
Injection volume: 25
Detector: UV 254

CHROMATOGRAM
Retention time: 3.6
Internal standard: 4'-nitroacetanilide (12.4)
Limit of detection: 3 μg/mL

OTHER SUBSTANCES
Extracted: cefamandole, cefotaxime, cefoxitin, cephapirin, chloramphenicol
Simultaneous: acetaminophen, N-acetylprocainamide, cefaclor, cephalexin, cephalothin, cimetidine, miconazole, moxalactam, procainamide, sulfamethoxazole, theophylline, tobramycin, vancomycin

KEY WORDS
serum

REFERENCE
Danzer,L.A. Liquid-chromatographic determination of cephalosporins and chloramphenicol in serum, *Clin.Chem.*, **1983**, *29*, 856–858.

SAMPLE
Matrix: blood
Sample preparation: 300 μL Plasma + 300 μL IS in ice-cold MeOH:100 mM pH 5.2 sodium acetate 70:30, vortex for 30 s, let stand at -20° for 10 min, centrifuge at 1500 g for 10 min, inject a 10 μL aliquot.

HPLC VARIABLES
Guard column: 4 × 4 10 μm C18
Column: 300 × 4 10 μm μBondapak C18
Mobile phase: MeCN:MeOH:100 mM sodium acetate 13.44:0.56:86, pH 5.2
Flow rate: 2.5
Injection volume: 10
Detector: UV 254

CHROMATOGRAM
Retention time: 5
Internal standard: cefoxitin (4)
Limit of detection: 200 ng/mL

KEY WORDS
plasma

REFERENCE
Signs,S.A.; File,T.M.; Tan,J.S. High-pressure liquid chromatographic method for analysis of cephalosporins, *Antimicrob.Agents Chemother.*, **1984**, *26*, 652–655.

SAMPLE
Matrix: blood
Sample preparation: Condition a 1 mL Bond-Elut C18 SPE cartridge with 2 mL MeOH and 2 mL 8.5% phosphoric acid. Condition an NH2 SPE cartridge with 1 mL hexane. 500 μL Plasma + 25 μL 8.5% phosphoric acid + 250 μL 1 mg/mL coumarin-3-carboxylic acid in water, add to the C18 SPE cartridge, wash with 500 μL water, wash with 1 mL 8.5% phosphoric acid, wash with 5% MeOH:8.5% phosphoric acid 20:1, elute with 1 mL MeOH: 8.5% phosphoric acid 60:40 into the NH2 SPE cartridge. Wash the NH2 SPE cartridge

with 1 mL hexane, wash with 1 mL MeCN, elute with 1 mL water:10% ammonium sulfate 95:5, inject a 20 μL aliquot of the eluate.

HPLC VARIABLES

Column: 250 × 4.6 C18
Mobile phase: Water:2 mM tetramethylammonium hydroxide in MeOH:acetic acid 60:40:0.5
Flow rate: 0.8
Injection volume: 20
Detector: UV 262

CHROMATOGRAM

Retention time: 10
Internal standard: coumarin-3-carboxylic acid (13)

OTHER SUBSTANCES

Extracted: ceftizoxime, cefaclor, cephalexin

KEY WORDS

plasma; SPE

REFERENCE

Moore,C.M.; Sato,K.; Hattori,H.; Katsumata,Y. Improved HPLC method for the determination of cephalosporins in human plasma and a new solid-phase extraction procedure for cefazolin and ceftizoxime [letter], *Clin.Chim.Acta*, **1990**, *190*, 121–123.

SAMPLE

Matrix: blood, urine
Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18
Column: 250 × 4.6 5 μm Symmetry C8 (Waters)
Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A:B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.
Column temperature: 30
Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)
Injection volume: 10-30
Detector: UV 200.5

CHROMATOGRAM

Retention time: 8.423

KEY WORDS

whole blood

REFERENCE

Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, *763*, 149–163.

SAMPLE

Matrix: bulk, formulations
Sample preparation: Dissolve in water to a concentration of 40 μg/mL, inject a 20 μL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:water:acetic acid 30:70:0.1
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM

Retention time: 11
Limit of quantitation: 1600 ng/mL

OTHER SUBSTANCES

Simultaneous: impurities, cefadroxil, cephapirin, ceftizoxime, cefaclor, cefotaxime, cephalexin, cefoxitin, cephradine, cefoperazone, cefamandole, cephalothin, cefamandole nafate

REFERENCE

Ting,S. Reverse-phase liquid chromatographic analysis of cephalosporins, *J.Assoc.Off.Anal.Chem.*, **1988**, *71*, 1123–1130.

SAMPLE

Matrix: cells
Sample preparation: 100 μL Cell suspension + 100 μL cefoperazone solution + 100 μL Hanks balanced salt solution, sonicate 30 min, add 800 μL MeCN, centrifuge at 13000 g for 5 min, remove supernatant. Dry supernatant under air, dissolve in 100 μL mobile phase, inject 75 μL.

HPLC VARIABLES

Column: μBondapak C18
Mobile phase: MeCN:50 mM pH 5.09 KH_2PO_4 10:90
Flow rate: 1
Injection volume: 75
Detector: UV 254

CHROMATOGRAM

Retention time: 11
Internal standard: Vancomycin
Limit of detection: 100-1000 ng/mL

REFERENCE

Darouiche,R.O.; Hamill,R.J. Antibiotic penetration of and bactericidal activity within endothelial cells, *Antimicrob.Agents Chemother.*, **1994**, *38*, 1059–1064.

SAMPLE

Matrix: formulations
Sample preparation: Inject a 20 μL aliquot.

HPLC VARIABLES
Column: 150 × 3.9 5 μm NovaPak phenyl
Mobile phase: MeCN:MeOH:0.5% phosphoric acid with 0.7% triethylamine 10:20:70
Flow rate: 1.5
Injection volume: 20
Detector: UV 322

CHROMATOGRAM
Retention time: 5.5

OTHER SUBSTANCES
Noninterfering: meperidine

KEY WORDS
injections; 5% dextrose; stability-indicating

REFERENCE
Lee,D.K.T.; Wong,C.-Y.; Wang,D.-P. Stability of cefazolin sodium and meperidine hydrochloride, *Am.J.Health-Syst.Pharm.*, **1996**, *53*, 1608–1610.

SAMPLE
Matrix: formulations
Sample preparation: Dilute with water (if necessary), inject a 20 μL aliquot.

HPLC VARIABLES
Column: 300 × 4 μBondapak phenyl
Mobile phase: MeOH:water 30:70 containing 10 mM ammonium acetate
Flow rate: 1.3
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 5.5

OTHER SUBSTANCES
Simultaneous: cephalothin

KEY WORDS
saline; 5% dextrose; stability-indicating

REFERENCE
Das Gupta,V.; Stewart,K.R. Quantitation of carbenicillin disodium, cefazolin sodium, cephalothin sodium, nafcillin sodium, and ticarcillin disodium by high-pressure liquid chromatography, *J.Pharm.Sci.*, **1980**, *69*, 1264–1267.

SAMPLE
Matrix: formulations
Sample preparation: Dilute with water, inject an aliquot.

HPLC VARIABLES
Column: 150 × 3.9 5 μm Nova Pak C18
Mobile phase: MeOH:5 mM pH 7.5 phosphate buffer 20:80
Flow rate: 1
Detector: UV 254

CHROMATOGRAM
Retention time: 2.7

OTHER SUBSTANCES
Simultaneous: degradation products, cefoxitin

KEY WORDS
injections; water; stability-indicating

REFERENCE
Stiles,M.L.; Tu,Y.H.; Allen,L.V.,Jr. Stability of cefazolin sodium, cefoxitin sodium, ceftazidime, and penicillin G sodium in portable pump reservoirs, *Am.J.Hosp.Pharm.*, **1989**, *46*, 1408–1412.

SAMPLE
Matrix: formulations
Sample preparation: Add theophylline (2 mg/mL), inject a 10 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:acetic acid:water 10:1:89, adjusted to pH 4 with 5 M NaOH
Flow rate: 1.5
Injection volume: 10
Detector: UV 293 (?)

CHROMATOGRAM
Retention time: 8.5
Internal standard: theophylline (4.1)

OTHER SUBSTANCES
Simultaneous: ceftazidime

KEY WORDS
injections; stability-indicating; 5% dextrose

REFERENCE
Bosso,J.A.; Prince,R.A.; Fox,J.L. Compatibility of ondansetron hydrochloride with fluconazole, ceftazidime, aztreonam, and cefazolin sodium under simulated Y-site conditions, *Am.J.Hosp.Pharm.*, **1994**, *51*, 389–391.

SAMPLE
Matrix: formulations
Sample preparation: Dilute with 5% dextrose (if necessary), inject a 20 μL aliquot.

HPLC VARIABLES
Column: Nova Pak C18
Mobile phase: MeOH:100 mM $(NH_4)_2HPO_4$ 20:80, pH 7.80
Flow rate: 1
Injection volume: 20
Detector: UV 322

CHROMATOGRAM
Retention time: 3.3

OTHER SUBSTANCES
Simultaneous: famotidine

KEY WORDS
injections; 5% dextrose; stability-indicating

REFERENCE

Wang,D.-P.; Chang,L.-C.; Wong,C.-Y.; Lee,D.K.T. Stability of cefazolin sodium-famotidine admixture, *Am.J.Hosp.Pharm.*, **1994**, *51*, 2205–2209.

SAMPLE

Matrix: solutions
Sample preparation: Dissolve sample in a mobile phase to concentration of about 1 mg/mL, inject a 10 μL aliquot.

HPLC VARIABLES

Column: 250 × 4.6 5 μm β-CyD (Advanced Separation Technologies Inc., USA)
Mobile phase: MeOH:buffer 42:58 (Buffer was 5 mM tetraethylammonium acetate adjusted to pH 3.6 with glacial acetic acid.)
Column temperature: 30
Flow rate: 0.8
Injection volume: 10
Detector: UV 230

CHROMATOGRAM

Retention time: 54

OTHER SUBSTANCES

Also analyzed: 7-ACA, 7-ADCA, cefaclor, cefaloridine, cefoperazone, cefotaxime, ceftazidime, cephalosporin C

REFERENCE

Tsou,T.-L.; Wu,J.-R.; Wang,T.-M. The effects of separation of cephalosporins by HPLC with β-cyclodextrin bonded stationary phase, *J.Liq.Chromatogr.Rel.Technol.*, **1996**, *19*, 1081–1095.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4 OmniPac PCX-500 (Dionex)
Mobile phase: Gradient. A was MeCN:90 mM perchloric acid 13.5:86.5. B was MeCN:300 mM perchloric acid 45:55. A:B from 100:0 to 0:100 over 7 min, maintain at 0:100.
Flow rate: 1
Detector: UV 254

CHROMATOGRAM

Retention time: 7

OTHER SUBSTANCES

Simultaneous: 7-aminocephalosproranic acid, cefadroxil, cefotaxime, cephalexin, cephaloridine, cephalosporin C, cephalothin, cephapirin, D-hydroxyphenylglycine

REFERENCE

Slingsby,R.W.; Rey,M. Determination of pharmaceuticals by multi-phase chromatography: Combined reversed phase and ion exchange in one column, *J.Liq.Chromatogr.*, **1990**, *13*, 107–134.

SAMPLE

Matrix: solutions
Sample preparation: Inject 100 μL onto column A with mobile phase A, after 3 min backflush the contents of column A onto column B with mobile phase B, elute column B with mobile phase B, monitor the effluent from column B.

HPLC VARIABLES
Column: A 30 × 0.3 5 μm ODS C18 (Nomura); B 150 × 0.3 5 μm ODS C18 (Nomura)
Mobile phase: A 10 mM ammonium acetate adjusted to pH 5 with acetic acid; B MeOH: water:acetic acid 40:60:0.5
Flow rate: A 0.1; B 0.004
Injection volume: 100
Detector: UV 262

CHROMATOGRAM
Retention time: 8.50
Limit of detection: 2 ng/mL

OTHER SUBSTANCES
Simultaneous: cefaclor, cephaloridine, ceftizoxime

KEY WORDS
microbore; column-switching

REFERENCE
Moore,C.M.; Sato,K.; Katsumata,Y. High-performance liquid chromatographic determination of cephalosporin antibiotics using 0.3 mm I.D. columns, *J.Chromatogr.*, **1991**, *539*, 215–220.

SAMPLE
Matrix: solutions
Sample preparation: Separate buffer containing drug from human serum albumin by centrifuging at 37° at 700 g for 3 min using a Micropartition System MPS-1 (Amicon) unit, inject a 10-20 μL aliquot of the ultrafiltrate.

HPLC VARIABLES
Guard column: C18/Corasil (Waters)
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:10 mM ammonium acetate 15:85
Flow rate: 1.5
Injection volume: 10-20
Detector: UV 270

OTHER SUBSTANCES
Also analyzed: cefpiramide, cefmenoxime, cefbuperazone, cefoxitin, cefotiam, cephaloridine

REFERENCE
Terasaki,T.; Nouda,H.; Tsuji,A. Relationship between lipophilicity and binding affinity with human serum albumin for penicillin and cephem antibiotics, *J.Pharmacobiodyn.*, **1992**, *15*, 99–106.

SAMPLE
Matrix: solutions
Sample preparation: Prepare a 2.5-5 μg/mL solution, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 80 × 4.6 3.65 μm Zorbax Rx-SIL (similar to Zorbax SB-C8 (Mac-Mod Analytical))
Mobile phase: MeCN:0.1% trifluoroacetic acid 20:80
Flow rate: 1
Injection volume: 20
Detector: UV 272

CHROMATOGRAM
Retention time: k' 2.9

REFERENCE

Kirkland,K.M.; McCombs,D.A.; Kirkland,J.J. Rapid, high-resolution high-performance liquid chromatographic analysis of antibiotics, *J.Chromatogr.A*, **1994**, *660*, 327–337.

SAMPLE

Matrix: solutions

Sample preparation: Inject a 10 μL aliquot.

HPLC VARIABLES

Column: 220 × 4.6 Spheri 5 ODS-224

Mobile phase: 100 mM sodium dodecyl sulfate, pH 3.00

Flow rate: 1

Injection volume: 10

Detector: UV 260

CHROMATOGRAM

Retention time: 2

OTHER SUBSTANCES

Simultaneous: cephalothin, cephaloridine, cephalexin, cephradine, 7-aminocephalorospanic acid, 7-aminodesacetoxycephalosporanic acid

REFERENCE

Garcia Pinto,C.; Pérez Pavón,J.L.; Moreno Cordero,B. Micellar liquid chromatography of zwitterions: Retention mechanism of cephalosporins, *Analyst*, **1995**, *120*, 53–62.

SAMPLE

Matrix: solutions

Sample preparation: Inject a 50-200 μL aliquot of a solution in pH 7.4 Tyrode's buffer.

HPLC VARIABLES

Column: 125 × 4 5 μm LiChrospher 60 RP-select B

Mobile phase: MeCN:10 mM pH 4 sodium acetate 12:88

Flow rate: 0.6

Injection volume: 50-200

Detector: UV 270

OTHER SUBSTANCES

Also analyzed: theophylline

KEY WORDS

buffer

REFERENCE

Saitoh,H.; Aungst,B.J. Possible involvement of multiple P-glycoprotein-mediated efflux systems in the transport of verapamil and other organic cations across rat intestine, *Pharm.Res.*, **1995**, *12*, 1304–1310.

SAMPLE

Matrix: tissue

Sample preparation: Condition a 100 mg Sep-Pak SPE cartridge with 5 mL MeOH and 5 mL water. Homogenize tissue with 4 (liver, lung) or 29 (spleen) volumes of water (Thomas tissue grinder series 3431-D70). 1 mL Homogenate + 20 μL 500 μg/mL cephalexin + 50 μL 8.5% phosphoric acid, vortex for 30 s, centrifuge at 2000 g for 5 min, add to the SPE cartridge, wash with 3 mL water, elute with 2 mL MeOH:water 60:40, inject a 10 μL aliquot of the eluate.

HPLC VARIABLES

Guard column: Nova-Pak C18 guard column
Column: 250 × 4.6 5 μm Econosphere C18
Mobile phase: MeOH:20 mM NaH_2PO_4 23:77, pH 5.0
Flow rate: 1
Injection volume: 100
Detector: UV 270

CHROMATOGRAM

Retention time: 9.5
Internal standard: cephalexin (11.5)
Limit of quantitation: 500 ng/g (spleen), 100 ng/g (liver, lung)

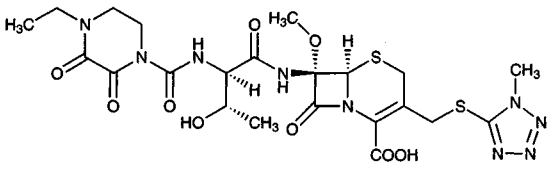
KEY WORDS

rat; liver; spleen; lung; SPE

REFERENCE

Liang,D.; Chow,D.; White,C. High-performance liquid chromatographic assay of cefazolin in rat tissues, *J.Chromatogr.B*, **1994**, *656*, 460–465.

Cefbuperazone



Molecular formula: $C_{22}H_{29}N_9O_9S_2$
Molecular weight: 627.66
CAS Registry No.: 76610-84-9
Merck Index: 1968
Lednicer No.: 4 189

SAMPLE

Matrix: solutions
Sample preparation: Separate buffer containing drug from human serum albumin by centrifuging at 37° at 700 g for 3 min using a Micropartition System MPS-1 (Amicon) unit, inject a 10-20 μL aliquot of the ultrafiltrate.

HPLC VARIABLES

Guard column: C18/Corasil (Waters)
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:10 mM ammonium acetate 15:85
Flow rate: 1.5
Injection volume: 10-20
Detector: UV 270

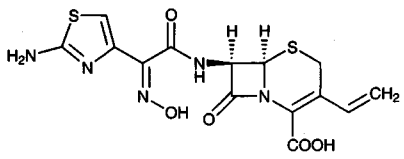
OTHER SUBSTANCES

Also analyzed: cefpiramide, cefazolin, cefmenoxime, cefoxitin, cefotiam, cephaloridine

REFERENCE

Terasaki,T.; Nouda,H.; Tsuji,A. Relationship between lipophilicity and binding affinity with human serum albumin for penicillin and cephem antibiotics, *J.Pharmacobiodyn.*, **1992**, *15*, 99–106.

Cefdinir



Molecular formula: $C_{14}H_{13}N_5O_5S_2$
Molecular weight: 395.42
CAS Registry No.: 91832-40-5
Merck Index: 1971

SAMPLE

Matrix: blood
Sample preparation: Mix plasma with an equal volume of MeCN, mix with 3.5 volumes of dichloromethane, inject a 20 μL aliquot of the aqueous phase.

HPLC VARIABLES

Column: 250 × 4.6 5 μm Spherisorb ODS1
Mobile phase: MeCN:water:perchloric acid:triethylamine 8:90.6:0.7:0.7
Flow rate: 1
Injection volume: 20
Detector: UV 284

CHROMATOGRAM

Retention time: 11
Limit of quantitation: 2 μg/mL

OTHER SUBSTANCES

Also analyzed: captopril, quinapril

KEY WORDS

plasma; pharmacokinetics; rat

REFERENCE

Jacolot,A.; Tod,M.; Petitjean,O. Pharmacokinetic interaction between cefdinir and two angiotensin-converting enzyme inhibitors in rats, *Antimicrob.Agents Chemother.*, **1996**, *40*, 979–982.

SAMPLE

Matrix: solutions
Sample preparation: Dissolve 200 mg cefdinir in 5 mL 2% sodium bicarbonate in water. Dilute 500 μL solution to 200 mL (100 μg/mL) with each 100 mM pH 3-8 phosphate buffer. For pH 9 studies dissolve 20 mg cefdinir in 200 mL 100 mM carbonate buffer. Inject an aliquot.

HPLC VARIABLES

Column: 75 × 4.6 TSK-gel ODS-80 TM (TOSOH, Japan)
Mobile phase: MeOH:dioxane:33 mM citric acid adjusted to pH 2.0 with 10% phosphoric acid 4:1:36 (Caution ! Dioxane is a carcinogen!)
Flow rate: 1.5
Detector: UV 254

CHROMATOGRAM

Retention time: 6.5

OTHER SUBSTANCES

Simultaneous: degradation products

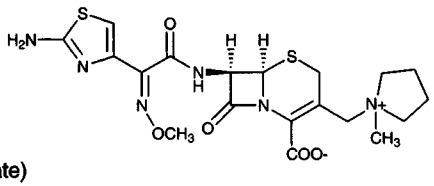
KEY WORDS

details for degradation of cefdinir in paper

REFERENCE

Okamoto,Y.; Kiriyama,K.; Namiki,Y.; Matsushita,J.; Fujioka,M.; Yasuda,T. Degradation kinetics and isomerization of cefdinir, a new oral cephalosporin, in aqueous solution. 1, *J.Pharm.Sci.*, **1996**, *85*, 976–983.

Cefepime



Molecular formula: $C_{19}H_{24}N_6O_5S_2$

Molecular weight: 480.57

CAS Registry No.: 88040-23-7, 123171-59-5 (HCl monohydrate)

Merck Index: 1973

SAMPLE

Matrix: blister fluid, blood

Sample preparation: Serum. 0.5 mL Serum + 2.5 mL MeCN, vortex, centrifuge at 3000 g for 5 min. Remove the supernatant and add it to 5 mL dichloromethane, vortex, inject a 10 μL aliquot of the aqueous layer. Blister fluid. Inject directly.

HPLC VARIABLES

Column: μBondapak C18

Mobile phase: MeCN:100 mM phosphate buffer 7:93

Flow rate: 2

Injection volume: 10

Detector: UV 229

CHROMATOGRAM

Retention time: 5

KEY WORDS

serum; pharmacokinetics

REFERENCE

Kalman,D.; Barriere,S.L.; Johnson,B.L.,Jr. Pharmacokinetic disposition and bactericidal activities of cefepime, ceftazidime, and cefoperazone in serum and blister fluid, *Antimicrob.Agents Chemother.*, **1992**, *36*, 453–457.

SAMPLE

Matrix: blood

Sample preparation: 500 μL Serum + 500 μL MeCN, vortex, gently shake by rotation at 20 rpm for 10 min. and centrifuge at 1000 g for 10 min. Remove the supernatant, add 3.2 mL dichloromethane, shake by rotation at 20 rpm for 10 min. and centrifuge at 1000 g for 10 min. Inject a 5 μL aliquot of the upper aqueous layer.

HPLC VARIABLES

Column: 75 × 4.6 3 μm Ultrasphere XL-ODS

Mobile phase: MeCN:20 mM ammonium acetate adjusted to pH 4 with glacial acetic acid 7:93

Flow rate: 1

Injection volume: 5

Detector: UV 254

CHROMATOGRAM

Retention time: 1.8-2.2

Limit of detection: 100 ng/mL

Limit of quantitation: 500 ng/mL

OTHER SUBSTANCES

Noninterfering: amikacin, amoxicillin, ampicillin, calcium folinate, cephalothin cefixime, cefotaxime, ceftazidime, ciprofloxacin, clavulanic acid, cloxacillin, erythromycin, fosfomycin, fusidic acid, gentamycin, hydroxyitraconazole, imipenem, itraconazole, kanamycin, latamoxef, lincomycin, mezlocillin, neomycin, netilmycin, ofloxacin, pefloxacin, penicillin G, piperacillin, pristinamycin, rifampicin, roxithromycin, sulbactam, sulfamethoxazole, tazobactam, teicoplanin, tetracycline, ticarcillin, tobramycin, trimethoprim, vancomycin

KEY WORDS

serum; pharmacokinetics

REFERENCE

Elkhaïli,H.; Linger,L.; Monteil,H.; Jehl,F. High-performance liquid chromatographic assay for cefepime in serum, *J.Chromatogr.B*, **1997**, *690*, 181–188.

SAMPLE

Matrix: blood

Sample preparation: 500 µL Serum + 900 µL MeCN:water:trichloroacetic acid 50:40:1.5 (v/v/w) + 1.5 mL dichloromethane, vortex for 10 s, centrifuge at 1200 g for 20 min, inject a 50 µL aliquot of the supernatant.

HPLC VARIABLES

Column: 5 µm C18
Mobile phase: MeCN:3.5% glycine adjusted to pH 9.5 with NaOH 5:95
Flow rate: 1
Injection volume: 50
Detector: UV 280 (from ref. 6)

CHROMATOGRAM

Limit of detection: 40 ng/mL

KEY WORDS

serum; dog

REFERENCE

Stampley,A.R.; Brown,M.P.; Gronwall,R.R.; Castro,L.; Stone,H.W. Serum concentrations of cefepime (BMY-28142), a broad-spectrum cephalosporin, in dogs, *Cornell.Vet.*, **1992**, *82*, 69–77.

SAMPLE

Matrix: blood, urine

Sample preparation: Plasma. 500 µL Plasma + 100 µL 100 µg/mL cefadroxil + 300 µL 5% trichloroacetic acid + 500 µL MeCN + 1.5 mL dichloromethane, vortex for 10 s, centrifuge at 500-600 g at 5° for 10 min, inject a 25 µL aliquot of the aqueous supernatant. Urine. Dilute urine three-fold with 200 mM pH 4.5 sodium acetate buffer, add 100 µL 1500 µg/mL ceftazidime, vortex for 30 s, inject a 10 µL aliquot.

HPLC VARIABLES

Guard column: 23 × 4 37-50 µm Corasil C18
Column: 150 × 4 Nova-Pak (plasma) or 100 × 9.4 Partisil 5 ODS-3RAC C18 (urine)
Mobile phase: MeCN:5 mM 1-octanesulfonic acid 12:88 (plasma) or MeOH:10 mM sodium dodecyl sulfate adjusted to pH 3.0 with glacial acetic acid:5% trichloroacetic acid:850 mM phosphoric acid:THF 49.7:40.4:3.9:0.7:5.3 (urine)
Flow rate: 1 (plasma), 2.8 (urine)
Injection volume: 10-25
Detector: UV 280

CHROMATOGRAM

Retention time: 7 (plasma), 7.5 (urine)

Internal standard: cefadroxil (10) (plasma), ceftazidime (10) (urine)
Limit of quantitation: 2000 ng/mL (urine), 100 ng/mL (plasma)

KEY WORDS
plasma; rat; pharmacokinetics

REFERENCE
Barbhaiya,R.H.; Forgue,S.T.; Shyu,W.C.; Papp,E.A.; Pittman,K.A. High-pressure liquid chromatographic analysis of BMY-28142 in plasma and urine, *Antimicrob.Agents Chemother.*, **1987**, *31*, 55–59.

SAMPLE
Matrix: blood, urine
Sample preparation: Plasma. 750 μL Plasma + 75 μL 200 mM pH 6.0 4-morpholineethanesulfonic acid buffer, mix, add 1.1 mL MeCN, centrifuge. Remove the supernatant and evaporate the MeCN under a stream of nitrogen, inject a 100 μL aliquot of the residue. Urine. Inject 100-200 μL urine directly.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeCN:water 5:95 containing 13 mM sodium pentanesulfonate, 11 mM sodium heptanesulfonate, and 10 mM sodium acetate, pH 3.0
Flow rate: 1
Injection volume: 100-200
Detector: Radioactivity

CHROMATOGRAM
Retention time: 45

KEY WORDS
plasma; rat; dog; radiolabeled

REFERENCE
Forgue,S.T.; Kari,P.; Barbhaiya,R. *N*-oxidation of *N*-methylpyrrolidine released *in vivo* from cefepime, *Drug Metab.Dispos.*, **1987**, *15*, 808–815.

SAMPLE
Matrix: blood, urine
Sample preparation: Serum. Precipitate protein in serum with MeCN and trichloroacetic acid, add dichloromethane, mix, centrifuge. Dilute aqueous supernatant with pH 4 sodium acetate buffer, inject an aliquot. Urine. Dilute urine with pH 4 sodium acetate buffer, inject an aliquot.

HPLC VARIABLES
Guard column: 30 × 4 30-40 μm Perisorb RP18
Column: 125 × 4 5 μm Nucleosil 5 C18
Mobile phase: MeCN:water:concentrated sulfuric acid 800:1197.5:2.5 containing 20 mM sodium dodecyl sulfate, adjust pH to 2.3 with concentrated sulfuric acid
Detector: UV 260

CHROMATOGRAM
Limit of detection: 1800 ng/mL, 270 ng/mL

KEY WORDS
serum

REFERENCE

Bächer,K.; Schaeffer,M.; Lode,H.; Nord,C.E.; Borner,K.; Koeppe,P. Multiple dose pharmacokinetics, safety, and effects on faecal microflora, of cefepime in healthy volunteers, *J.Antimicrob.Chemother.*, **1992**, *30*, 365–375.

SAMPLE

Matrix: cecal contents

Sample preparation: Dilute cecal contents in 2 mL phosphate buffered saline, centrifuge at 1500 g for 10 min. 500 μL Sample + 300 μL 5% trichloroacetic acid + 500 μL MeCN + 1.5 mL dichloromethane, vortex for 10 s, centrifuge at 500-600 g at 5° for 10 min, inject a 20 μL aliquot of the upper aqueous phase.

HPLC VARIABLES

Column: 100 × 3 5 μm Hypersil ODS

Mobile phase: MeCN: 5 mM pH 5.5 acetate buffer 0.7:99.3

Flow rate: 1

Injection volume: 20

Detector: UV 280

KEY WORDS

mouse; pharmacokinetics

REFERENCE

van Ogtrop,M.L.; Guiot,H.F.L.; Mattie,H.; van Furth,R. Modulation of the intestinal flora of mice by parenteral treatment with broad-spectrum cephalosporins, *Antimicrob.Agents Chemother.*, **1991**, *35*, 976–982.

SAMPLE

Matrix: cell suspensions

Sample preparation: Filter (0.45 μm).

HPLC VARIABLES

Column: 150 × 4.6 5 μm Ultrasphere IP ion pair

Mobile phase: MeOH:100 mM sodium perchlorate adjusted to pH 2.5 with concentrated sulfuric acid 30:70

Flow rate: 1

Injection volume: 20

Detector: UV 254

CHROMATOGRAM

Retention time: 3.6

OTHER SUBSTANCES

Also analyzed: cefpirome, carumonam (UV 295), ceftriaxone, cefotaxime

REFERENCE

Bellido,F.; Pechère,J.-C.; Hancock,R.E.W. Novel method for measurement of outer membrane permeability to new β-lactams in intact *Enterobacter cloacae* cells, *Antimicrob.Agents Chemother.*, **1991**, *35*, 68–72.

SAMPLE

Matrix: tissue

Sample preparation: Homogenize muscle with three volumes phosphate buffered saline (Polytron, level 3) for 2 min, centrifuge at 1300 g for 10 min. 125 μL Supernatant + 100 μL 40 μg/mL cefadroxil in water + 800 μL MeCN, vortex for 30 s, centrifuge at 1600 g for 5 min. Remove the supernatant and evaporate it to dryness under a stream of nitrogen, reconstitute the residue in 125 μL mobile phase, inject a 50 μL aliquot.

HPLC VARIABLES

Column: 4 μm Novapak C18
Mobile phase: MeCN:5 mm sodium heptanesulfonic acid 9:91, adjust pH to 3.33 with glacial acetic acid
Flow rate: 2
Injection volume: 50
Detector: UV 280

CHROMATOGRAM

Retention time: 4
Internal standard: cefadroxil (6.7)
Limit of detection: 800 ng/g, 200 ng/mL

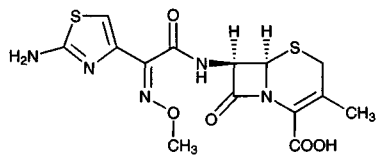
KEY WORDS

mouse; muscle

REFERENCE

Darouiche,R.; Musher,D.; Hamill,R.; Ou,C.; Rognerud,C. Cephalosporin penetration into soft tissue of paralyzed limbs, *Antimicrob.Agents Chemother.*, **1989**, *33*, 1326–1328.

Cefetamet



Molecular formula: $C_{14}H_{15}N_5O_5S_2$
Molecular weight: 397.44
CAS Registry No.: 65052-63-3
Merck Index: 1974
Lednicer No.: 4 184

SAMPLE

Matrix: blood
Sample preparation: 500 μL Plasma + 500 μL 500 mM perchloric acid, vortex for 5 s, let stand for 15 min, centrifuge at 1500 g for 5 min, inject a 50 μL aliquot of the supernatant.

HPLC VARIABLES

Column: 125 × 4 Nucleosil 5 C18
Mobile phase: MeCN:100 mM pH 6.5 phosphate buffer 40:60
Flow rate: 1
Injection volume: 50 (cefetamet pivoxyl)
Detector: UV 265

CHROMATOGRAM

Retention time: 5.6
Limit of detection: 200-400 ng/mL
Limit of quantitation: 500 ng/mL

KEY WORDS

plasma; human; dog; rat

REFERENCE

Wyss,R.; Bucheli,F. Determination of cefetamet and its orally active ester, cefetamet pivoxyl, in biological fluids by high-performance liquid chromatography, *J.Chromatogr.*, **1988**, *430*, 81–92.

SAMPLE

Matrix: blood, urine
Sample preparation: Plasma. 500 μL Plasma + 500 μL 500 mM perchloric acid, vortex for 5 s, let stand for 15 min, centrifuge at 1500 g for 5 min, inject a 30 μL aliquot of the supernatant. Urine. Dilute 500 μL urine to 25 mL with water, vortex for 5 s, inject a 30 μL aliquot.

HPLC VARIABLES

Column: 125 × 4 5 μm Spherisorb ODS 1
Mobile phase: MeCN:4 mM perchloric acid 17:83 (plasma) or 15:85 (urine)
Flow rate: 1
Injection volume: 30
Detector: UV 265

CHROMATOGRAM

Retention time: 8 (plasma), 11 (urine)
Limit of quantitation: 20 μg/mL (urine), 200 ng/mL (plasma)

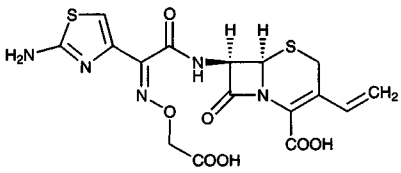
KEY WORDS

plasma; human; dog; rat

REFERENCE

Wyss,R.; Bucheli,F. Determination of cefetamet and its orally active ester, cefetamet pivoxyl, in biological fluids by high-performance liquid chromatography, *J.Chromatogr.*, **1988**, *430*, 81–92.

Cefixime



Molecular formula: $C_{16}H_{15}N_5O_7S_2$
Molecular weight: 453.46
CAS Registry No.: 79350-37-1
Merck Index: 1975
Lednicer No.: 4 184,185

SAMPLE

Matrix: blood, urine

Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES

Guard column: 20 mm long Symmetry C18
Column: 250 × 4.6 5 μm Symmetry C8 (Waters)
Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A:B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.
Column temperature: 30
Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)
Injection volume: 10-30
Detector: UV 287.8

CHROMATOGRAM

Retention time: 4.823

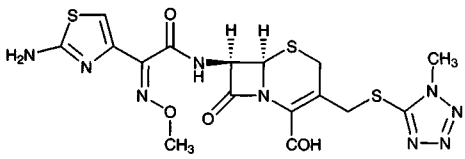
KEY WORDS

whole blood

REFERENCE

Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, *763*, 149–163.

Cefmenoxime



Molecular formula: $C_{16}H_{17}N_9O_5S_3$
Molecular weight: 511.57
CAS Registry No.: 65085-01-0, 75738-58-8 (HCl)
Merck Index: 1976
Lednicer No.: 4 187

SAMPLE

Matrix: bile, blood, urine
Sample preparation: Serum. 0.5 mL serum + 0.5 mL MeCN mix in 7 mL tube on vortex mixer; shake by rotation (20 rpm) 10 min; centrifuge 10 min 1000 g; transfer supernatant to another tube, add 7 aliquots dichloromethane; equilibrate 10 min; shake by rotation (20 rpm) 10 min; centrifuge 10 min 1000 g; inject aliquot of upper aqueous layer. Urine. Centrifuge urine and dilute 1:20. Bile. Centrifuge bile and dilute 1:10.

HPLC VARIABLES

Column: 150 × 4.6 5 μm Ultrasphere ODS
Mobile phase: 12:88 MeCN:20 mM ammonium acetate adjusted to pH 5 with glacial acetic acid
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM

Retention time: 7.6
Limit of detection: 200 ng/mL

OTHER SUBSTANCES

Also analyzed: ampicillin, azlocillin, aztreonam, cefoperazone, cefsulodin, cefotaxime, ceftazidime, ceftriaxone, cloxacillin, desacetylcefotaxime, mezlocillin, penicillin G, piperacillin, ticarcillin

KEY WORDS

serum

REFERENCE

Jehl,F.; Birckel,P.; Monteil,H. Hospital routine analysis of penicillins, third-generation cephalosporins and aztreonam by conventional and high-speed high-performance liquid chromatography, *J.Chromatogr.*, **1987**, *413*, 109–119.

SAMPLE

Matrix: blood
Sample preparation: 1 mL Plasma + 200 μL 7.8 μg/mL IS in 4% sodium dodecyl sulfate, centrifuge using an Amicon Model 25 filter (MW cutoff 25000) at 450 g for 20 min, inject a 90 μL aliquot of the ultrafiltrate.

HPLC VARIABLES

Column: 300 × 4 10 μm μBondapak C18
Mobile phase: MeCN:200 mM pH 5.30 acetate buffer 13:87
Flow rate: 2
Injection volume: 90
Detector: UV 254

CHROMATOGRAM

Retention time: 6

Internal standard: p-nitrobenzoic acid (3) or p-anisic acid (7)
Limit of detection: 50 ng/mL

KEY WORDS
plasma; ultrafiltrate

REFERENCE
Granneman,G.R.; Sennello,L.T. A very precise high-performance liquid chromatographic procedure for the determination of cefmenoxime, a new cephalosporin antibiotic, in plasma, *J.Chromatogr.*, **1982**, *229*, 149–157.

SAMPLE
Matrix: blood, CSF
Sample preparation: Serum. 1 mL Serum + 1 mL MeCN, vortex, centrifuge at 5000 rpm for 10 min. Remove the supernatant and add it to 4 mL dichloromethane, vortex, centrifuge. Remove the aqueous supernatant and inject an aliquot. CSF. Inject CSF directly.

HPLC VARIABLES
Column: endcapped 5 μm Lichrospher RP 18
Mobile phase: MeCN:2% acetic acid 25:75
Flow rate: 1
Injection volume: 4.61
Detector: UV 254

CHROMATOGRAM
Retention time: 20

KEY WORDS
serum

REFERENCE
Condomines,M.; Mallet,M.N.; Albanese,J.; Gouin,F.; De Micco,P. A rapid high-performance liquid chromatography method for determining β-lactam antibiotics in biological fluids and tissues, *Chemioterapia*, **1987**, *6*, 251–253.

SAMPLE
Matrix: blood, urine
Sample preparation: 500 μL Serum or urine + 1 mL 10 (serum) or 100 (urine) μg/mL cefuroxime in MeOH, shake on a microthermomixer for 30 s, filter (urine samples only), centrifuge at 3000 rpm for 3 min, filter (0.5 μm) the supernatant, dilute with an equal volume of water, inject a 50 μL aliquot.

HPLC VARIABLES
Guard column: 10 × 4 Nucleosil 5C18
Column: 150 × 4 Nucleosil 5C18
Mobile phase: MeCN:water:acetic acid 10:50:1
Flow rate: 0.7
Injection volume: 50
Detector: UV 254

CHROMATOGRAM
Retention time: 8
Internal standard: cefuroxime (11)
Limit of detection: 2000 ng/mL (urine), 50 ng/mL (serum)

KEY WORDS
serum

REFERENCE
Itakura,K.; Mitani,M.; Aoki,I.; Usui,Y. High performance liquid chromatographic assay of cefsulodin, cefotiam and cefmenoxime in serum and urine, *Chem.Pharm.Bull.(Tokyo)*, **1982**, *30*, 622–627.

SAMPLE
Matrix: blood, urine
Sample preparation: Plasma. 200 μL Plasma + 200 μL 0.8 μg/mL p-anisic acid in MeCN, vortex for 10 sa, centrifuge at 700 g for 10 min. Remove the supernatant and evaporate it to 100 μL under a stream of nitrogen, vortex briefly, inject a 25-50 μL aliquot. Urine. Dilute, add p-anisic acid (200 μg/mL), inject an aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak phenyl
Mobile phase: MeCN:0.2% phosphoric acid 14:86
Flow rate: 2
Injection volume: 25-50
Detector: UV 254

CHROMATOGRAM
Retention time: 10
Internal standard: p-anisic acid (14)
Limit of detection: 5000 ng/mL (urine), 200 ng/mL (plasma)

KEY WORDS
plasma; pharmacokinetics

REFERENCE
Noonan,I.A.; Gambertoglio,J.G.; Barriere,S.L.; Conte,J.E.,Jr.; Lin,E.T. High-performance liquid chromatographic determination of cefmenoxime (AB-50912) in human plasma and urine, *J.Chromatogr.*, **1983**, *273*, 458–463.

SAMPLE
Matrix: blood, urine
Sample preparation: Dilute urine 100-fold. 500 μL Serum, plasma, or diluted urine + 100 μL 24 μg/mL p-anisic acid in water, vortex for 30 s, add 100 μL perchloric acid, vortex for 30 s, centrifuge at 2000 rpm for 15 min, inject a 25 μL aliquot of the supernatant.

HPLC VARIABLES
Column: μBondapak CN
Mobile phase: pH 3.8 Buffer prepared from 109 mL 13.61 g/L sodium acetate trihydrate solution + 891 mL 5.75 mL/L acetic acid solution. (After each batch run MeOH through column at 3 mL/min for 10 min.)
Flow rate: 2.5
Injection volume: 25
Detector: UV 254

CHROMATOGRAM
Retention time: 3.4
Internal standard: p-anisic acid (5.6)
Limit of detection: 500 ng/mL

OTHER SUBSTANCES
Noninterfering: acetaminophen, cimetidine, diazepam, digoxin, dopamine, furosemide, heparin, hydralazine, hydrochlorothiazide, hydrocortisone, hydromorphone, isosorbide dinitrate, methyldopa, nitroglycerin, quinidine, theophylline, tobramycin

KEY WORDS
serum; plasma; pharmacokinetics

REFERENCE

Reitberg,D.P.; Schentag,J.J. Liquid-chromatographic assay of cefmenoxime in serum and urine, *Clin.Chem.*, **1983**, *29*, 1415–1418.

SAMPLE

Matrix: solutions

Sample preparation: Separate buffer containing drug from human serum albumin by centrifuging at 37° at 700 g for 3 min using a Micropartition System MPS-1 (Amicon) unit, inject a 10-20 μL aliquot of the ultrafiltrate.

HPLC VARIABLES

Guard column: C18/Corasil (Waters)
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:10 mM ammonium acetate 15:85
Flow rate: 1.5
Injection volume: 10-20
Detector: UV 260

OTHER SUBSTANCES

Also analyzed: cefpiramide, cefazolin, cefbuperazone, cefoxitin, cefotiam, cephaloridine

REFERENCE

Terasaki,T.; Nouda,H.; Tsuji,A. Relationship between lipophilicity and binding affinity with human serum albumin for penicillin and cephem antibiotics, *J.Pharmacobiodyn.*, **1992**, *15*, 99–106.

SAMPLE

Matrix: surface wipes

Sample preparation: Swab 100 × 100 mm surface with 1% pH 6 phosphate buffer (total volume 10 mL), remove excess liquid with a second swab, vortex swabs for 45 s, filter (0.45 μm polycarbonate), inject a 100 μL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeCN:water:acetic acid 15:100:1
Flow rate: 2
Injection volume: 100
Detector: UV 254

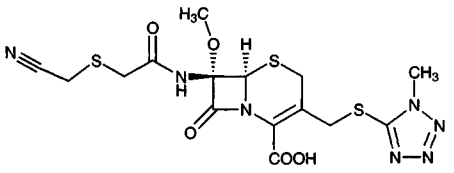
CHROMATOGRAM

Retention time: 7.1
Limit of quantitation: 100 ng/mL

REFERENCE

Gorski,R.J.; Plasz,A.C.; Elrod,L.J.; Yoder,J.; White,L.B. Determination of cefsulodin, cefmenoxime, and cefadroxil as residues on surfaces, *Pharm.Res.*, **1991**, *8*, 1525–1527.

Cefmetazole



Molecular formula: $C_{15}H_{17}N_7O_5S_3$
Molecular weight: 471.54
CAS Registry No.: 56796-20-4, 56796-39-5 (sodium salt)
Merck Index: 1977
Lednicer No.: 4 190

SAMPLE

Matrix: blood, urine
Sample preparation: Blood. Add 500 μL MeOH to 500 μL serum, vortex for 30 s, centrifuge at 10000 g for 15 min, inject a 50 μL aliquot of the supernatant. Urine. Add 4.5 mL 50 mM pH 6.0 potassium phosphate buffer to 500 μL urine, mix vigorously for 15 s, filter 1 mL of this solution (0.45 μm filter, Millex HA, Millipore), inject a 50 μL aliquot of the filtrate.

HPLC VARIABLES

Guard column: 30 × 4.6 5 μm Nucleosil-5 C18
Column: 150 × 4.6 5 μm Nucleosil-5 C18
Mobile phase: A MeCN:100 mM pH 6.0 potassium phosphate buffer 13:87; B MeCN:100 mM pH 6.0 potassium phosphate buffer 20:80 containing 0.1 mM hexadecyltrimethylammonium
Flow rate: 1
Injection volume: 50
Detector: UV 272

CHROMATOGRAM

Retention time: 7.4 (A), 7.7 (B)
Limit of quantitation: 200 ng/mL (serum), 2 μg/mL (urine)

KEY WORDS

serum

REFERENCE

García-Glez,J.C.; Méndez,R.; Martín-Villacorta,J. Quantitative determination of semisynthetic cephamycins in human serum and urine by ion-exchange, reversed-phase and ion-pair chromatography, *J.Chromatogr.A*, **1998**, *812*, 197–204.

SAMPLE

Matrix: blood, urine
Sample preparation: Mix, deproteinize, add barbital, vortex, centrifuge, inject an aliquot.

HPLC VARIABLES

Column: C18
Mobile phase: MeCN:20 mM pH 5.4 citrate buffer 12:88
Flow rate: 1.5
Detector: UV 254

CHROMATOGRAM

Internal standard: barbital
Limit of quantitation: 15000 ng/mL (urine), 2000 ng/mL (plasma)

KEY WORDS

plasma; pharmacokinetics

REFERENCE

Borin,M.T.; Peters,G.R.; Smith,T.C. Pharmacokinetics and dose proportionality of cefmetazole in healthy young and elderly volunteers, *Antimicrob.Agents Chemother.*, **1990**, *34*, 1944–1948.

SAMPLE

Matrix: formulations
Sample preparation: Inject a 20 μL aliquot.

HPLC VARIABLES

Column: Nova Pak C18
Mobile phase: MeCN:0.1% acetic acid:10 mM pH 7.8 $(NH_4)H_2PO_4$ 10:23:74
Flow rate: 1
Injection volume: 20
Detector: UV 300

CHROMATOGRAM

Retention time: 8.1

OTHER SUBSTANCES

Simultaneous: famotidine
Noninterfering: degradation products

KEY WORDS

stability-indicating; injections; 5% dextrose

REFERENCE

Lee,D.K.T.; Wong,C.-Y.; Wang,D.-P.; Chang,L.-C.; Wu,K.-H. Stability of cefmetazole sodium and famotidine, *Am.J.Health-Syst.Pharm.*, **1996**, *53*, 432–442.

SAMPLE

Matrix: milk
Sample preparation: Mix 10 mL milk with 2 mL 100 mM tetraethylammonium chloride, add 40 mL MeCN slowly with continual stirring, let stand for 10 min, decant the supernatant through a plug of glass wool. Collect 40 mL filtrate, add 2 mL buffer, evaporate to 1-2 mL under reduced pressure at 40-50°, dilute to 4 mL with water, filter (0.45 μm PVDF). Inject a 2 mL aliquot onto a 150 × 4.6 5 μm Supelcosil LC-18 column, elute with MeCN:10 mM KH_2PO_4 0:100 for 3 min, to 60:40 over 37 min at 1 mL/min, collect a 1.5-2 mL aliquot containing the compound (ca. 19.7 min), evaporate to <1 mL under reduced pressure, make up to 1 mL with water, inject an aliquot. (Prepare the buffer by mixing 10 mM KH_2PO_4 and 10 mM Na_2HPO_4 in a 5:1 ratio, pH 6.)

HPLC VARIABLES

Column: 150 × 4.6 5 μm Supelcosil LC-18
Mobile phase: MeCN:buffer 18:82 (Buffer was 20 mM phosphoric acid containing 10 mM sodium decanesulfonate.)
Flow rate: 1
Injection volume: 200
Detector: UV 260

REFERENCE

Moats,W.A.; Romanowski,R.D. Multiresidue determination of β-lactam antibiotics in milk and tissues with the aid of high-performance liquid chromatographic fractionation for clean up, *J.Chromatogr.A*, **1998**, *812*, 237–247.

SAMPLE

Matrix: solutions

Sample preparation: Separate buffer containing drug from human serum albumin by centrifuging at 37° at 700 g for 3 min using a Micropartition System MPS-1 (Amicon) unit, inject a 10-20 μL aliquot of the ultrafiltrate.

HPLC VARIABLES

Guard column: C18/Corasil (Waters)
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:10 mM ammonium acetate + 10 mM tetrabutylammonium bromide + 1% acetic acid 25:75
Flow rate: 1.5
Injection volume: 10-20
Detector: UV 270

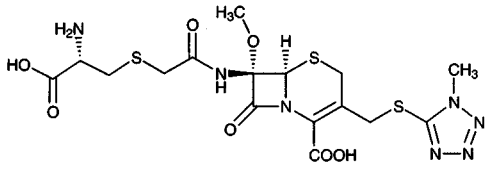
OTHER SUBSTANCES

Also analyzed: cefpiramide, cefoperazone, cefmenoxime, ceftezole

REFERENCE

Terasaki,T.; Nouda,H.; Tsuji,A. Relationship between lipophilicity and binding affinity with human serum albumin for penicillin and cephem antibiotics, *J.Pharmacobiodyn.*, **1992**, *15*, 99–106.

Cefminox



Molecular formula: $C_{16}H_{21}N_7O_7S_3$
Molecular weight: 519.58
CAS Registry No.: 84305-41-9, 75481-73-1
Merck Index: 1978

SAMPLE

Matrix: blood, urine

Sample preparation: Blood. Add 500 μL MeOH to 500 μL serum, vortex for 30 s, centrifuge at 10000 g for 15 min, inject a 50 μL aliquot of the supernatant. Urine. Add 4.5 mL 50 mM pH 6.0 potassium phosphate buffer to 500 μL urine, mix vigorously for 15 s, filter 1 mL of this solution (0.45 μm filter, Millex HA, Millipore), inject a 50 μL aliquot of the filtrate.

HPLC VARIABLES

Guard column: 30 × 4.6 5 μm Nucleosil-5 C18
Column: 150 × 4.6 5 μm Nucleosil-5 C18
Mobile phase: A MeCN:100 mM pH 6.0 potassium phosphate buffer 3:97; B MeCN:100 mM pH 6.0 potassium phosphate buffer 7:93 containing 0.1 mM hexadecyltrimethylammonium
Flow rate: 1
Injection volume: 50
Detector: UV 272

CHROMATOGRAM

Retention time: 7.8 (A), 6.2 (B)
Limit of quantitation: 200 ng/mL (serum), 2 μg/mL (urine)

KEY WORDS

serum

REFERENCE

García-Glez,J.C.; Méndez,R.; Martín-Villacorta,J. Quantitative determination of semisynthetic cephamycins in human serum and urine by ion-exchange, reversed-phase and ion-pair chromatography, *J.Chromatogr.A*, **1998**, *812*, 197–204.

SAMPLE

Matrix: solutions

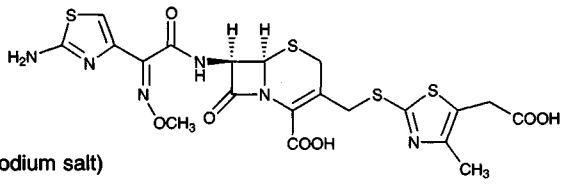
HPLC VARIABLES

Column: 100 × 4 5 μm ODS-Hypersil
Mobile phase: MeOH:water:o-phosphoric acid 20:80:1
Flow rate: 2
Detector: UV 270

REFERENCE

Soriano,F.; Edwards,R.; Greenwood,D. Comparative susceptibility of cefminox and cefoxitin to β-lactamases of *Bacteroides* spp., *J.Antimicrob.Chemother.*, **1991**, *28*, 55–60.

Cefodizime



Molecular formula: $C_{20}H_{20}N_6O_7S_4$
Molecular weight: 584.68
CAS Registry No.: 69739-16-8, 86329-79-5 (disodium salt)
Merck Index: 1979

SAMPLE

Matrix: bile, blood, feces, tissue, urine

Sample preparation: Plasma, serum. 500 µL Plasma or serum + 2 mL ice-cold 2.5 µg/mL 3,5-dinitrobenzoic acid in MeOH, vortex for 30 s, place in an ice-bath for 30 min, centrifuge at 2000 g in a refrigerated centrifuge for 15 min. Remove the supernatant and concentrate it under nitrogen to 500 µL, inject a 20 µL aliquot. Urine. 1 mL Centrifuged urine + 4 mL 25 µg/mL 3,5-dinitrobenzoic acid in 100 mM pH 7.0 phosphate buffer, mix well, filter (0.45 µm), inject a 20 µL aliquot. Bile. 500 µL Centrifuged bile + 1 mL 15 µg/mL 3,5-dinitrobenzoic acid in 100 mM pH 7.0 phosphate buffer, stir, filter (0.45 µm), inject a 20 µL aliquot. Feces. 1 g Homogenized feces + 4 mL ice-cold EtOH:1% pH 6.0 phosphate buffer 2:1, shake vigorously for 5 min, centrifuge in a refrigerated centrifuge at 2000 g for 10 min, filter (0.45 µm) the supernatant. 500 µL Supernatant + 500 µL 10 µg/mL 3,5-dinitrobenzoic acid in 100 mM pH 7.0 phosphate buffer, mix, inject a 20 µL aliquot. Tissue. Homogenize 0.1-1 g visceral tissue with 4 mL 100 mM pH 7.0 phosphate buffer, centrifuge at 5° at 2000 g for 15 min. 500 µL Supernatant + 2 mL ice-cold 5 µg/mL 3,5-dinitrobenzoic acid in MeOH, vortex vigorously, let stand at 5° for 30 min, centrifuge at 5° at 2000 g for 15 min. Remove the supernatant and concentrate it to 500 µL under nitrogen, inject a 20 µL aliquot. (Prepare 100 mM pH 7.0 phosphate buffer with 35.81 $Na_2HPO_4.12H_2O$ and 13.61 g KH_2PO_4 in 1 L water, adjust pH with NaOH or phosphoric acid if necessary. Prepare 1% pH 6.0 phosphate buffer with 6.0 g $Na_2HPO_4.12H_2O$ and 7.0 g KH_2PO_4 in 1 L water, adjust pH with NaOH or phosphoric acid if necessary.)

HPLC VARIABLES

Column: 100 × 8 4 µm Radial-Pak NOVA C18
Mobile phase: MeCN:water:acetic acid 20:80:2 containing 5 mM sodium 1-hexanesulfonate
Flow rate: 2
Injection volume: 20
Detector: UV 264

CHROMATOGRAM

Retention time: 7.7
Internal standard: 3,5-dinitrobenzoic acid (10.2)
Limit of detection: 300 ng/mL (bile), 500 ng/mL (urine), 500 ng/g (feces, tissue), 100 ng/mL (plasma)

OTHER SUBSTANCES

Simultaneous: piperacillin, cefmetazole, cefotetan, cefoperazone, cefotaxime

KEY WORDS

plasma; serum; human; rat; liver; spleen; lung; kidney; heart; brain; intestine; stomach; thymus

REFERENCE

Marunaka,T.; Matsushima,E.; Maniwa,M. Determination of cefodizime in biological materials by high-performance liquid chromatography, *J.Chromatogr.*, **1987**, *420*, 329–339.

SAMPLE

Matrix: blood

Sample preparation: Dilute serum with an equal volume of water, inject a 20 μL aliquot onto column A, elute column A to waste with MeOH:10 mM pH 7.0 phosphate buffer 5:95 at 0.3 mL/min, after 1.3 min elute the contents of column A onto column B with mobile phase A or B, elute with mobile phase A or B, monitor the effluent from column B.

HPLC VARIABLES

Column: A 50 × 2.1 40 μm Supelclean LC-NH_2; B 150 × 4.6 3 μm Supelcosil LC-18
Mobile phase: A MeCN:MeOH:10 mM pH 7.0 phosphate buffer 15:20:65 containing 5 mM tetrabutylammonium hydrogen sulfate; B MeOH:10 mM pH 7.0 phosphate buffer 30:70 containing 5 mM tetrabutylammonium hydrogen sulfate
Injection volume: 20
Detector: UV 267

CHROMATOGRAM

Retention time: 5.7 (mobile phase A), 6.8 (mobile phase B)
Limit of detection: 500-2000 ng/mL

OTHER SUBSTANCES

Extracted: cefamandole, cefazolin, cefoperazone, cefoxitin, ceftizoxime, ceftriaxone, cefuroxime, cephaloridine, cephalothin
Noninterfering: acetaminophen, acyclovir, digoxin, fluconazole, ranitidine, teicoplanin, theophylline, vancomycin

KEY WORDS

column-switching; serum

REFERENCE

Bompadre,S.; Ferrante,L.; Leone,L. On-line solid-phase extraction of cephalosporins, *J.Chromatogr.A*, **1998**, *812*, 191–196.

SAMPLE

Matrix: blood
Sample preparation: Dilute plasma with three volumes of mobile phase A, inject a 100 μL aliquot on to column A and elute to waste with mobile phase A, after 1 min elute the contents of column A on to column B (already equilibrated with mobile phase A) with mobile phase A, after 2.5 min remove column A from the circuit and elute column B with mobile phase B, monitor the effluent from column B. Before the next injection equilibrate columns A and B with mobile phase A.

HPLC VARIABLES

Column: A 50 × 4 40 μm CN silica; B 200 × 4 5 μm HP ODS (Hewlett-Packard)
Mobile phase: A MeCN:5 mM sodium 1-heptanesulfonate:acetic acid 10:88:2; B MeCN:5 mM sodium 1-heptanesulfonate:acetic acid 27:71:2
Flow rate: 1
Injection volume: 100
Detector: UV 263

CHROMATOGRAM

Retention time: 7.6
Limit of detection: 100 ng/mL

KEY WORDS

column-switching; plasma; pharmacokinetics

REFERENCE

Bompadre,S.; Ferrante,L.; Leone,L.; de Martinis,M.; Ginaldi,L.; Quaglino,D. Determination of cefodizime in human plasma by high-performance liquid chromatography with column-switching, *J.Liq.Chromatogr.*, **1995**, *18*, 2895–2909.

SAMPLE

Matrix: blood, urine

Sample preparation: Plasma. 250 μL Plasma + uvitic acid + 500 μL MeOH, centrifuge, add to SAX-modified silica SPE cartridge, wash with water, elute with 500 μL 1 M NaCl, add 500 μL water to the eluate, inject a 500 μL aliquot. Urine. 100 μL Urine + uvitic acid + 900 μL 100 mM pH 5.0 acetate buffer, add to SAX-modified silica SPE cartridge, wash with water, elute with 500 μL 1 M NaCl, add 500 μL water to the eluate, inject a 500 μL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 μm μBondapak C18

Mobile phase: MeCN:100 mM pH 4.0 citrate buffer 14:86 (plasma) or 13:87 (urine)

Flow rate: 1.5

Injection volume: 500

Detector: UV 300

CHROMATOGRAM

Retention time: 13

Internal standard: uvitic acid (9.5)

Limit of quantitation: 1 μg/mL (urine), 20 ng/mL (plasma)

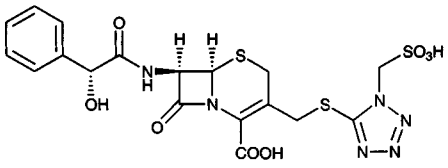
KEY WORDS

plasma; SPE; pharmacokinetics

REFERENCE

Lenfant,B.; Namour,F.; Logeais,C.; Coussediere,D.; Rivault,O.; Bryskier,A.; Surjus,A. Pharmacokinetics of cefodizime following single doses of 0.5, 1.0, 2.0, and 3.0 grams administered intravenously to healthy volunteers, *Antimicrob.Agents Chemother.*, **1995**, *39*, 2037–2041.

Cefonicid



Molecular formula: $C_{18}H_{18}N_6O_8S_3$
Molecular weight: 542.57
CAS Registry No.: 61270-58-4, 61270-78-8 (disodium salt), 71420-79-6 (monosodium salt)
Merck Index: 1980
Lednicer No.: 3 213

SAMPLE

Matrix: blood, urine
Sample preparation: Plasma, serum. 200 μL Plasma or serum + 400 μL 2.1 μg/mL cephalothin in MeCN:water 5:1, vortex for 10 s, centrifuge at 1500 g for 10 min. Remove the supernatant and evaporate it to 200 μL under nitrogen, vortex, inject a 5-20 μL aliquot. Urine. 200 μL Urine + 400 μL 8.3 μg/mL cephalothin in MeCN:water 5:1, vortex for 10 s, centrifuge at 1500 g for 10 min. Remove the supernatant and inject a 5-20 μL aliquot.

HPLC VARIABLES

Column: 300 × 3.9 10 μm μBondapak phenyl
Mobile phase: MeCN:water:phosphoric acid:tetrabutylammonium hydrogen sulfate 25:74.6:0.1:0.3
Flow rate: 2
Injection volume: 5-20
Detector: UV 254

CHROMATOGRAM

Retention time: 13.5
Internal standard: cephalothin (11.5)
Limit of detection: 500 ng/mL

KEY WORDS

plasma; serum

REFERENCE

Phelps,R.; Zurlinden,E.; Conte,J.E.,Jr.; Lin,E. High-performance liquid chromatographic determination of cefonicid in human plasma, serum and urine, *J.Chromatogr.*, **1986**, *375*, 111–118.

SAMPLE

Matrix: blood, urine
Sample preparation: 100 μL Plasma or urine + 100 μL (500 μL for urine) 10 μg/mL cefoperazone in 100 mM pH 5 ammonium acetate buffer, vortex for 15 s, centrifuge at 8700 g for 2 min, inject a 20-200 μL aliquot onto column A with mobile phase A, wash with mobile phase A for 2 min then backflush contents of column A onto column B with mobile phase B, monitor effluent from column B.

HPLC VARIABLES

Column: A 20 × 4 35-50 μm C18 Corasil; B 20 × 4 35-50 μm C18 Corasil + 100 × 8 10 μm Radial-Pak μBondapak C18
Mobile phase: A water:triethylamine 1000:4, adjusted to pH 3.0 with orthophosphoric acid; B MeCN:water:triethylamine 750:250:4, adjusted to pH 3.0 with orthophosphoric acid
Flow rate: A 2; B 3.8
Injection volume: 20-200
Detector: UV 270

CHROMATOGRAM
Retention time: 2.33
Internal standard: cefoperazone (3.66)
Limit of detection: 250 ng/mL

OTHER SUBSTANCES
Simultaneous: ceftazidime, ceftriaxone, cefotaxime, cephaloridine, ceforanide, moxalactam, cephalothin
Noninterfering: cefotiam, cefadroxil
Interfering: cefazolin

KEY WORDS
plasma; column-switching

REFERENCE
Demotes-Mainard,F.; Vinçon,G.; Jarry,C.; Necciari,J.; Albin,H. Micromethod for the determination of cefpiramide in human plasma and urine by high-performance liquid chromatography using automated column switching, *J.Chromatogr.*, **1987**, *419*, 388–395.

SAMPLE
Matrix: solutions
Sample preparation: Add 100 μL solution to 1 mL 0.5 mg/mL cefoperazone in water, vortex for 15 s, inject a 10 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 10 μm Alltech C8
Mobile phase: MeCN:20 mM sodium acetate 30:70 containing 1.7 g/L tetrabutylammonium hydrogen sulfate, pH adjusted to 6.0 with 1 M NaOH
Flow rate: 1.5
Injection volume: 10
Detector: UV 254

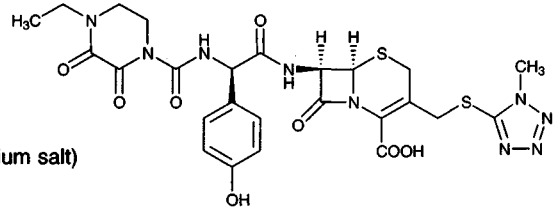
CHROMATOGRAM
Retention time: 8.5
Internal standard: cefoperazone (5.3)

OTHER SUBSTANCES
Simultaneous: degradation products

KEY WORDS
5% dextrose; saline

REFERENCE
Marble,D.A.; Bosso,J.A.; Townsend,R.J. Compatibility of clindamycin phosphate with aztreonam in polypropylene syringes and with cefoperazone sodium, cefonicid sodium, and cefuroxime sodium in partial-fill glass bottles, *Drug Intell.Clin.Pharm.*, **1988**, *22*, 54–57.

Cefoperazone



Molecular formula: $C_{25}H_{27}N_9O_8S_2$
Molecular weight: 645.68
CAS Registry No.: 62893-19-0, 62893-20-3 (sodium salt)
Merck Index: 1981
Lednicer No.: 4 185, 188-190

SAMPLE

Matrix: bile, blood, urine
Sample preparation: Serum. 0.5 mL serum + 0.5 mL MeCN mix in 7 mL tube on vortex mixer; shake by rotation (20 rpm) 10 min; centrifuge 10 min 1000 g; transfer supernatant to another tube, add 7 aliquots dichloromethane; equilibrate 10 min; shake by rotation (20 rpm) 10 min; centrifuge 10 min 1000 g; inject aliquot of upper aqueous layer. Urine. Centrifuge urine and dilute 1:20. Bile. Centrifuge bile and dilute 1:10

HPLC VARIABLES

Column: 150 × 4.6 5 μm Ultrasphere ODS
Mobile phase: 20:80 MeCN:20 mM ammonium acetate adjusted to pH 5 with glacial acetic acid
Flow rate: 1
Injection volume: 20
Detector: UV 254

OTHER SUBSTANCES

Also analyzed: ampicillin, azlocillin, aztreonam, cefmenoxime, cefsulodin, cefotaxime, ceftazidime, ceftriaxone, cloxacillin, desacetylcefotaxime, mezlocillin, penicillin G, piperacillin, ticarcillin

KEY WORDS

serum

REFERENCE

Jehl,F.; Birckel,P.; Monteil,H. Hospital routine analysis of penicillins, third-generation cephalosporins and aztreonam by conventional and high-speed high-performance liquid chromatography, *J.Chromatogr.*, **1987**, *413*, 109–119.

SAMPLE

Matrix: bile, blood, urine
Sample preparation: Dilute bile and urine with water. 50 μL Plasma, diluted urine, or diluted bile + 50 μL 10% perchloric acid + 50 μL 3-butylxanthine in pH 7.4 phosphate buffer, mix, centrifuge at 15000 g for 10 min, inject an aliquot of the supernatant.

HPLC VARIABLES

Column: Cosmosil 5C18 (Nacalai Tesque)
Mobile phase: MeOH:30 mM pH 5.0 KH_2PO_4 20:80
Column temperature: 50
Flow rate: 1.5
Detector: UV 266

CHROMATOGRAM

Internal standard: 3-butylxanthine
Limit of detection: 100 ng/mL
Limit of quantitation: 200 ng/mL

KEY WORDS
rat; plasma; pharmacokinetics

REFERENCE
Haghgoo,S.; Hasegawa,T.; Nadai,M.; Wang,L.; Nabeshima,T.; Kato,N. Effect of a bacterial lipopolysaccharide on biliary excretion of a β-lactam antibiotic, cefoperazone, in rats, *Antimicrob.Agents Chemother.*, **1995**, *39*, 2258–2261.

SAMPLE
Matrix: blood
Sample preparation: Dilute serum with an equal volume of water, inject a 20 μL aliquot onto column A, elute column A to waste with MeOH:10 mM pH 7.0 phosphate buffer 5:95 at 0.3 mL/min, after 1.3 min elute the contents of column A onto column B with mobile phase A or B, elute with mobile phase A or B, monitor the effluent from column B.

HPLC VARIABLES
Column: A 50 × 2.1 40 μm Supelclean LC-NH_2; B 150 × 4.6 3 μm Supelcosil LC-18
Mobile phase: A MeCN:MeOH:10 mM pH 7.0 phosphate buffer 15:20:65 containing 5 mM tetrabutylammonium hydrogen sulfate; B MeOH:10 mM pH 7.0 phosphate buffer 30:70 containing 5 mM tetrabutylammonium hydrogen sulfate
Flow rate: 1
Injection volume: 20
Detector: UV 267

CHROMATOGRAM
Retention time: 6.0 (mobile phase A), 7.8 (mobile phase B)
Limit of detection: 500-2000 ng/mL

OTHER SUBSTANCES
Extracted: cefamandole, cefazolin, cefodizime, cefoxitin, ceftizoxime, ceftriaxone, cefuroxime, cephaloridine, cephalothin
Noninterfering: acetaminophen, acyclovir, digoxin, fluconazole, ranitidine, teicoplanin, theophylline, vancomycin

KEY WORDS
column-switching; serum

REFERENCE
Bompadre,S.; Ferrante,L.; Leone,L. On-line solid-phase extraction of cephalosporins, *J.Chromatogr.A*, **1998**, *812*, 191–196.

SAMPLE
Matrix: blood
Sample preparation: 300 μL Plasma + 300 μL IS in ice-cold MeOH:100 mM pH 5.2 sodium acetate 70:30, vortex for 30 s, let stand at -20° for 10 min, centrifuge at 1500 g for 10 min, inject a 10 μL aliquot.

HPLC VARIABLES
Guard column: 4 × 4 10 μm C18
Column: 300 × 4 10 μm μBondapak C18
Mobile phase: MeCN:MeOH:100 mM sodium acetate 18.24:0.76:81, pH 5.2
Flow rate: 2.5
Injection volume: 10
Detector: UV 254

CHROMATOGRAM
Retention time: 4

Internal standard: cephaloridine (3)
Limit of detection: 1000 ng/mL

KEY WORDS
plasma

REFERENCE
Signs,S.A.; File,T.M.; Tan,J.S. High-pressure liquid chromatographic method for analysis of cephalosporins, *Antimicrob.Agents Chemother.*, **1984**, *26*, 652–655.

SAMPLE
Matrix: blood
Sample preparation: 50 μL Plasma + 150 μL MeOH, vortex for 30 s, incubate at room temperature for 5 min, centrifuge at 1500 g for 10 min, inject a 20 μL aliquot of the supernatant.

HPLC VARIABLES
Guard column: 30 × 4.6 10 μm Develosil ODS-10
Column: 250 × 4.6 10 μm Develosil ODS-10
Mobile phase: MeOH:5 mM Na_2HPO_4 + 5 mM NaH_2PO_4 1:2
Flow rate: 1.2
Injection volume: 20
Detector: UV 265

KEY WORDS
plasma; human; rat

REFERENCE
Haginaka,J.; Wakai,J.; Yasuda,H.; Uno,T.; Nakagawa,T. High-performance liquid chromatographic assay of sulbactam using pre-column reaction with 1,2,4-triazole, *J.Chromatogr.*, **1985**, *341*, 115–122.

SAMPLE
Matrix: blood
Sample preparation: 500 μL Serum + 500 μL ice-cold 100 μg/mL ceftriaxone in MeOH: 100 mM pH 5.2 sodium acetate 70:30, vortex for 30 s, hold at -20° for 10 min, centrifuge at 1500 g for 10 min, inject 15 μL of supernatant.

HPLC VARIABLES
Guard column: 10 μm C18 Guard-PAK
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeCN:10 mM pH 7.5 phosphate buffer containing 10 mM hexadecyltrimethylammonium bromide 25:75
Flow rate: 1.3
Injection volume: 15
Detector: UV 254

CHROMATOGRAM
Internal standard: ceftriaxone
Limit of detection: 800 ng/mL

KEY WORDS
serum

REFERENCE
Deeter,R.G.; Weinstein,M.P.; Swanson,K.A.; Gross,J.S.; Bailey,L.C. Crossover assessment of serum bactericidal activity and pharmacokinetics of five broad-spectrum cephalosporins in the elderly, *Antimicrob.Agents Chemother.*, **1990**, *34*, 1007–1013.

SAMPLE
Matrix: blood
Sample preparation: 500 μL Plasma + 500 μL MeCN, mix vigorously on a Whirlmixer for 30 s, centrifuge at 1200 g for 5 min. Remove 400 μL of the supernatant and add it to 3 mL dichloromethane, centrifuge at 1200 g for 5 min, inject a 20 μL aliquot of the upper aqueous layer.

HPLC VARIABLES
Column: 100 × 3 5 μm Hypersil ODS
Mobile phase: MeCN:5 mM pH 5.5 acetate buffer 0.7:97.3
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Limit of detection: 200 ng/mL

OTHER SUBSTANCES
Also analyzed: ceftriaxone, ceftazidime, cefepime

KEY WORDS
plasma; mouse

REFERENCE
van Ogtrop,M.L.; Mattie,H.; Guiot,H.F.L.; van Strijen,E.; Hazekamp-van Dokkum,A.-M.; van Furth,R. Comparative study of the effects of four cephalosporins against Escherichia coli in vitro and in vivo, *Antimicrob.Agents Chemother.*, **1990**, *34*, 1932–1937.

SAMPLE
Matrix: blood, tissue
Sample preparation: Homogenize 1 g tissue with 10 mL pH 6.0 100 mM phosphate buffer (Polytron homogenizer), centrifuge at 3000 g for 10 min. 0.5 mL Serum or tissue homogenate supernatant + 2 mL MeCN, vortex for 1 min, centrifuge at 3000 g for 5 min. Remove the supernatant and add it to 5 mL dichloromethane, vortex , centrifuge at 3000 g for 5 min, inject a 50 μL aliquot of the upper layer.

HPLC VARIABLES
Column: 300 mm long μBondapak C18
Mobile phase: MeCN:100 mM sodium phosphate 16:84, adjust pH to 6.0
Flow rate: 2.5
Injection volume: 50
Detector: UV 254

CHROMATOGRAM
Retention time: 6

OTHER SUBSTANCES
Also analyzed: cefoxitin

KEY WORDS
serum

REFERENCE
Bawdon,R.E.; Hemsell,D.L.; Guss,S.P. Comparison of cefoperazone and cefoxitin concentrations in serum and pelvic tissue of abdominal hysterectomy patients, *Antimicrob.Agents Chemother.*, **1982**, *22*, 999–1003.

SAMPLE
Matrix: blood, urine
Sample preparation: Serum. 1 mL Serum + 1 mL MeOH, vortex for 30 s, allow to stand at room temperature for 10 min, if necessary dilute with water to give a cefoperazone concentration of 1-100 μg/mL, centrifuge at 1022 g for 10 min, inject a 20 μL aliquot. Urine. Centrifuge at 1022 g for 10 min, if necessary dilute with water to give a cefoperazone concentration of 1-100 μg/mL, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 10 μm μBondapak C18
Mobile phase: Gradient. A was 1.2 mM triethylamine and 42 mM acetic acid. B was MeCN: water 24:76 containing 1.2 mM triethylamine and 42 mM acetic acid. A:B from 75:25 to 60:40 using Waters Model 660 curve select-9 over 15 min then stay at 60:40.
Flow rate: 2
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 23
Limit of quantitation: 1000 ng/mL

OTHER SUBSTANCES
Simultaneous: degradation products, ampicillin, methicillin, penicillin G
Noninterfering: gentamicin, kanamycin, tobramycin

KEY WORDS
serum

REFERENCE
Dokladalova,J.; Quercia,G.T.; Stankewich,J.P. High-performance liquid chromatographic determination of cefoperazone in human serum and urine, *J.Chromatogr.*, **1983**, *276*, 129–137.

SAMPLE
Matrix: blood, urine
Sample preparation: 500 (?) μL Plasma or urine + 1500 (?) μL MeCN, vortex, centrifuge, inject a 20 μL aliquotof the supernatant.

HPLC VARIABLES
Column: Brownlee 10 μm RP-8
Mobile phase: MeCN:MeOH:20 mM pH 5 phosphate buffer 10:20:70
Flow rate: 2
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 4

OTHER SUBSTANCES
Also analyzed: ceftriaxone, cefaronide

KEY WORDS
plasma; sheep; pharmacokinetics

REFERENCE
Guerrini,V.H.; Filippich,L.J.; Cao,G.R.; English,P.B.; Bourne,D.W.A. Pharmacokinetics of cefaronide, ceftriaxone and cefoperazone in sheep, *J.Vet.Pharmacol.Ther.*, **1985**, *8*, 120–127.

SAMPLE

Matrix: blood, urine

Sample preparation: 100 μL Plasma or urine + 100 μL (500 μL for urine) 100 mM pH 5 ammonium acetate buffer, vortex for 15 s, centrifuge at 8700 g for 2 min, inject a 20-200 μL aliquot onto column A with mobile phase A, wash with mobile phase A for 2 min then backflush contents of column A onto column B with mobile phase B, monitor effluent from column B.

HPLC VARIABLES

Column: A 20 × 4 35-50 μm C18 Corasil; B 20 × 4 35-50 μm C18 Corasil + 100 × 8 10 μm Radial-Pak μBondapak C18

Mobile phase: A water:triethylamine 1000:4, adjusted to pH 3.0 with orthophosphoric acid; B MeCN:water:triethylamine 750:250:4, adjusted to pH 3.0 with orthophosphoric acid

Flow rate: A 2; B 3.8

Injection volume: 20-200

Detector: UV 270

CHROMATOGRAM

Retention time: 3.66

Internal standard: cefoperazone

Limit of detection: 250 ng/mL

OTHER SUBSTANCES

Simultaneous: cefonicid, ceftazidime, ceftriaxone, cefotaxime, cephaloridine, ceforanide, moxalactam, cephalothin

Noninterfering: cefotiam, cefadroxil

Interfering: cefazolin

KEY WORDS

plasma; column-switching; cefoperazone is IS

REFERENCE

Demotes-Mainard,F.; Vinçon,G.; Jarry,C.; Necciari,J.; Albin,H. Micromethod for the determination of cefpiramide in human plasma and urine by high-performance liquid chromatography using automated column switching, *J.Chromatogr.*, **1987**, *419*, 388–395.

SAMPLE

Matrix: blood, urine

Sample preparation: 1 mL Plasma, plasma water, or urine + 1 mL 10% trichloroacetic acid + 16 μg phenacetin, vortex for 1 min, centrifuge at 3000 rpm for 12 min, inject an aliquot of the supernatant.

HPLC VARIABLES

Column: Novapak C18

Mobile phase: MeOH:10 mM pH 5.5 KH_2PO_4 13:83 (sic)

Detector: UV 254

CHROMATOGRAM

Internal standard: phenacetin

Limit of quantitation: 1 μg/mL

KEY WORDS

plasma; plasma water; pharmacokinetics

REFERENCE

Hu,O.Y.-P.; Tang,H.-S.; Chang,C.-L. The influence of chronic lobular hepatitis on pharmacokinetics of cefoperazone—a novel galactose single-point method as a measure of residual liver function, *Biopharm.Drug Dispos.*, **1994**, *15*, 563–576.

SAMPLE
Matrix: blood, urine
Sample preparation: 1 mL Plasma or urine + 1 mL 10% trichloroacetic acid + 16 μg phenacetin, vortex for 1 min, centrifuge at 3000 rpm for 12 min, inject an aliquot of the supernatant.

HPLC VARIABLES
Column: Novapak C18
Mobile phase: MeOH:10 mM pH 5.5 KH_2PO_4 13:83
Detector: UV 254

CHROMATOGRAM
Internal standard: phenacetin
Limit of detection: 1000 ng/mL

KEY WORDS
plasma; pharmacokinetics

REFERENCE
Hu,O.Y.; Tang,H.S.; Chang,C.L. Novel galactose single point method as a measure of residual liver function: example of cefoperazone kinetics in patients with liver cirrhosis, *J.Clin.Pharmacol.*, **1995**, *35*, 250–258.

SAMPLE
Matrix: bulk, formulations
Sample preparation: Dissolve in water, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:water:acetic acid 30:70:0.1
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 18
Limit of quantitation: 1680 ng/mL

OTHER SUBSTANCES
Simultaneous: impurities, cefadroxil, cephapirin, ceftizoxime, cefaclor, cefotaxime, cephalexin, cefazolin, cefoxitin, cephradine, cefamandole, cephalothin, cefamandole nafate

REFERENCE
Ting,S. Reverse-phase liquid chromatographic analysis of cephalosporins, *J.Assoc.Off.Anal.Chem.*, **1988**, *71*, 1123–1130.

SAMPLE
Matrix: cecal contents
Sample preparation: Dilute cecal contents in 2 mL phosphate buffered saline, centrifuge at 1500 g for 10 min. 500 μL Sample + 500 μL MeCN, vortex for 30 s, centrifuge at 1200 g for 5 min. Remove 400 μL of the supernatant and add it to 3 mL dichloromethane, mix for 30 s, centrifuge at 1200 g for 5 min, inject a 20 μL aliquot of the upper aqueous phase.

HPLC VARIABLES
Column: 100 × 3 5 μm Hypersil ODS
Mobile phase: MeCN: 5 mM pH 5.5 acetate buffer 0.7:99.3
Flow rate: 1

Injection volume: 20
Detector: UV 254

OTHER SUBSTANCES
Also analyzed: ceftazidime, ceftriaxone

KEY WORDS
mouse; pharmacokinetics

REFERENCE
van Ogtrop,M.L.; Guiot,H.F.L.; Mattie,H.; van Furth,R. Modulation of the intestinal flora of mice by parenteral treatment with broad-spectrum cephalosporins, *Antimicrob.Agents Chemother.*, **1991**, *35*, 976–982.

SAMPLE
Matrix: cecal contents
Sample preparation: Weigh contents of cecum, dilute with 2 mL PBS, centrifuge at 1500 g for 10 min. Add a 500 μL aliquot of supernatant to 500 μL MeCN, mix on a whirlmixer for 30 s, centrifuge at 1200 g for 5 min. Remove 400 μL of the supernatant and add it to 3 mL dichloromethane, mix for 30 s, centrifuge at 1200 g for 5 min, inject a 20 μL aliquot of the upper aqueous phase.

HPLC VARIABLES
Column: 100 × 3 5 μm Hypersil ODS
Mobile phase: MeCN:5 mM pH 5.5 acetate buffer 0.7:99.3
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Limit of quantitation: 500 ng/mL

OTHER SUBSTANCES
Also analyzed: ceftazidime, ceftriaxone, cefepime

KEY WORDS
mouse

REFERENCE
van Ogtrop,M.L.; Guiot,H.F.L.; Mattie,H.; van Furth,R. Modulation of the intestinal flora of mice by parenteral treatment with broad-spectrum cephalosporins, *Antimicrob.Agents Chemother.*, **1991**, *35*, 976–982.

SAMPLE
Matrix: cells
Sample preparation: 100 μL Cell suspension + 100 μL cefoperazone solution + 100 μL Hanks balanced salt solution, sonicate 30 min, add 800 μL MeCN, centrifuge at 13000 g for 5 min, remove supernatant. Dry supernatant under air, dissolve in 100 μL mobile phase, inject 75 μL.

HPLC VARIABLES
Column: μBondapak C18
Mobile phase: MeCN:10 mM pH 5.2 ammonium acetate 15:85
Flow rate: 1
Injection volume: 75
Detector: UV 254

CHROMATOGRAM
Retention time: 14.8
Internal standard: Cefuroxime
Limit of detection: 100-1000 ng/mL

REFERENCE
Darouiche,R.O.; Hamill,R.J. Antibiotic penetration of and bactericidal activity within endothelial cells, *Antimicrob.Agents Chemother.*, **1994**, *38*, 1059–1064.

SAMPLE
Matrix: solutions
Sample preparation: Dissolve sample in mobile phase to a concentration of about 1 mg/mL, inject a 10 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm β-CyD (Advanced Separation Technologies Inc., USA)
Mobile phase: MeOH:buffer 42:58 (Buffer was 5 mM tetraethylammonium acetate, adjusted to pH 3.6 with glacial acetic acid.)
Column temperature: 30
Flow rate: 0.8
Injection volume: 10
Detector: UV 230

CHROMATOGRAM
Retention time: 47

OTHER SUBSTANCES
Also analyzed: 7-ACA, 7-ADCA, cefaclor, cefaloridine, cefazolin, cefotaxime, ceftazidime, cephalosporin C

REFERENCE
Tsou,T.-L.; Wu,J.-R.; Wang,T.-M. The effects of separation of cephalosporins by HPLC with β-cyclodextrin bonded stationary phase, *J.Liq.Chromatogr.Rel.Technol.*, **1996**, *19*, 1081–1095.

SAMPLE
Matrix: solutions

HPLC VARIABLES
Column: 250 × 4 LiChrosorb RP-18
Mobile phase: MeOH:pH 4.55 acetate buffer 23:77
Flow rate: 1.5
Injection volume: 10
Detector: UV 270

CHROMATOGRAM
Retention time: 5
Internal standard: diprophylline (10)

OTHER SUBSTANCES
Simultaneous: degradation products

REFERENCE
Jelinska,A.; Zajac,M. Effect of amino acids and amines on the stability of cefoperazone, *Pharmazie*, **1996**, *51*, 162–164.

SAMPLE
Matrix: solutions

Sample preparation: Add 100 μL solution to 1 mL 1 mg/mL aztreonam in water, vortex for 15 s, inject a 10 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 10 μm Alltech C8
Mobile phase: MeCN:10 mM acetate buffer 28:72 containing 1.7 g/L tetrabutylammonium hydrogen sulfate, pH adjusted to 3.5 with 5 M NaOH (The buffer was 240 mL 10 mM sodium acetate and 480 mL 10 mM acetic acid.)
Flow rate: 1.5
Injection volume: 10
Detector: UV 254

CHROMATOGRAM
Retention time: 10.4
Internal standard: aztreonam (6.2)

OTHER SUBSTANCES
Simultaneous: degradation products

KEY WORDS
5% dextrose; saline

REFERENCE
Marble,D.A.; Bosso,J.A.; Townsend,R.J. Compatibility of clindamycin phosphate with aztreonam in polypropylene syringes and with cefoperazone sodium, cefonicid sodium, and cefuroxime sodium in partial-fill glass bottles, *Drug Intell.Clin.Pharm.*, **1988**, *22*, 54–57.

SAMPLE
Matrix: solutions
Sample preparation: Separate buffer containing drug from human serum albumin by centrifuging at 37° at 700 g for 3 min using a Micropartition System MPS-1 (Amicon) unit, inject a 10-20 μL aliquot of the ultrafiltrate.

HPLC VARIABLES
Guard column: C18/Corasil (Waters)
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeCN:10 mM ammonium acetate 22:78
Flow rate: 1.5
Injection volume: 10-20
Detector: UV 230

OTHER SUBSTANCES
Also analyzed: penicillin G, methicillin, cephalothin

REFERENCE
Terasaki,T.; Nouda,H.; Tsuji,A. Relationship between lipophilicity and binding affinity with human serum albumin for penicillin and cephem antibiotics, *J.Pharmacobiodyn.*, **1992**, *15*, 99–106.

SAMPLE
Matrix: solutions

HPLC VARIABLES
Column: 125 × 4.6 Lichrospher 100 RP-18
Mobile phase: MeOH:2.5 mM pH 5.6 sodium phosphate buffer 18:80
Flow rate: 1
Injection volume: 20
Detector: UV 274

CHROMATOGRAM

Retention time: 9
Limit of detection: 60 nM

OTHER SUBSTANCES

Simultaneous: cefoxitin, ceftazidime, cefuroxime, cephalexin, cephradine

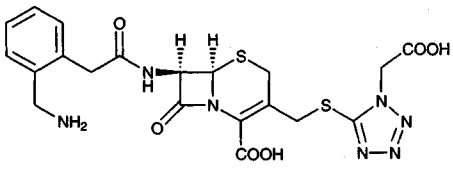
KEY WORDS

comparison with capillary electrophoresis

REFERENCE

Choi,O.-K.; Song,Y.-S. Determination of cefuroxim levels in human serum by micellar electrokinetic capillary chromatography with direct sample injection, *J.Pharm.Biomed.Anal.*, **1997**, *15*, 1265–1270.

Ceforanide



Molecular formula: $C_{20}H_{21}N_7O_6S_2$
Molecular weight: 519.56
CAS Registry No.: 60925-61-3
Merck Index: 1982
Lednicer No.: 3 214

SAMPLE

Matrix: blood

Sample preparation: 500 μL Plasma + 50 μL 1 mg/mL sulfacetamide + 100 μL 6% trichloroacetic acid, vortex for 1 min, add 1 mL MeCN, centrifuge at 2500 g for 10 min. Remove the supernatant and add it to 5 mL dichloromethane, vortex for 1 min, shake for 10 min, centrifuge, inject a 100 μL aliquot of the aqueous phase. Analyze for free ceforanide by centrifuging serum at 3000 rpm for 20 min through an Amicon micropartition system with YMT membranes, 200 μL ultrafiltrate + 20 μL 1 mg/mL sulfacetamide, inject a 100 μL aliquot.

HPLC VARIABLES

Column: 250 mm long C18 (Alltech)
Mobile phase: MeOH:100 mM pH 4.0 sodium acetate buffer 10:90
Flow rate: 2
Injection volume: 100
Detector: UV 254

CHROMATOGRAM

Internal standard: sulfacetamide

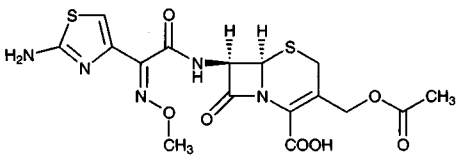
KEY WORDS

plasma; pharmacokinetics

REFERENCE

DiPiro,J.T.; Bayoumi,S.M.; Vallner,J.J.; Nesbit,R.R.; Gokhale,R.; Rissing,J.P. Intraoperative ceforanide pharmacokinetics and protein binding, *Antimicrob.Agents Chemother.*, **1985**, *27*, 487–490.

Cefotaxime



Molecular formula: $C_{16}H_{17}N_5O_7S_2$
Molecular weight: 455.47
CAS Registry No.: 63527-52-6, 64485-93-4 (sodium salt)
Merck Index: 1983
Lednicer No.: 3 216

SAMPLE

Matrix: aqueous humor, blood
Sample preparation: Aqueous humor. Inject a 10 μL aliquot directly. Plasma. Condition a 3 mL C18 SPE cartridge (Varian) with two 3 mL portions of MeCN and 3 mL buffer. Add 2 mL 625 ng/mL ciprofloxacin in buffer to 500 μL of plasma, mix, add to the SPE cartridge. Wash with 3 mL buffer. Remove moisture with vacuum (200 mbar) for 10 min. Elute with two 500 μL portions of MeCN:buffer 40:60. Vortex the eluate, inject a 10 μL aliquot. (Buffer was 100 mM Tris adjusted to pH 5.0 with HCl).

HPLC VARIABLES

Column: 300 × 4.6 5 μm endcapped ODS-Hypersil
Mobile phase: MeCN:DMF:10 mM NaH_2PO_4 15:6:79, adjusted to pH 3.0 with 85% phosphoric acid
Flow rate: 1
Injection volume: 10
Detector: UV 285

CHROMATOGRAM

Retention time: 6.7
Internal standard: ciprofloxacin (12.0)
Limit of detection: 80 ng/mL (aqueous humor), 310 ng/mL (plasma)

OTHER SUBSTANCES

Extracted: metabolites, ofloxacin

KEY WORDS

plasma; SPE

REFERENCE

Kraemer,H.-J.; Gehrke,R.; Breithaupt,A.; Breithaupt,H. Simultaneous quantification of cefotaxime, desacetylcefotaxime, ofloxacine and ciprofloxacine in ocular aqueous humor and in plasma by high-performance liquid chromatography, *J.Chromatogr.B*, **1997**, *700*, 147–153.

SAMPLE

Matrix: blood, tissue, urine
Sample preparation: Plasma. 50 μL Plasma + 50 μL MeCN, mix for 30 s, centrifuge at 5000 g for 15 min. Inject an aliquot of the supernatant. Urine. Mix 200 μL MeCN and 100 μL urine for 30 s, centrifuge at 5000 g for 15 min. Inject an aliquot. Tissue. Weight out finely chopped tissue and suspend it in 200 μL water, sonicate for 60 s. Add 200 μL MeCN, vortex for 30 s, centrifuge at 10000 g for 15 min. Inject an aliquot.

HPLC VARIABLES

Guard column: 15 × 3.2 7 μm Newguard C18 (Alltech)
Column: 250 × 4.6 5 μm Alltima C18 (Alltech)
Mobile phase: MeCN:50 mM pH 5.0 sodium dihydrogen phosphate 10:90
Flow rate: 1.0
Detector: UV 215

CHROMATOGRAM
Retention time: 11.6
Internal standard: cefotaxime

OTHER SUBSTANCES
Extracted: ampicillin

KEY WORDS
cefotaxime is IS; plasma; muscle; rat

REFERENCE
Cross,S.E.; Thompson,M.J.; Roberts,M.S. Distribution of systemically administered ampicillin, benzylpenicillin, and flucloxacillin in excisional wounds in diabetic and normal rats and effects of local topical vasodilator treatment, *Antimicrob.Agents Chemother.*, **1996**, *40*, 1703–1710.

SAMPLE
Matrix: solutions
Sample preparation: Dissolve sample in mobile phase to a concentration of about 1 mg/mL, inject a 10 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm β-CyD (Advanced Separation Technologies Inc., USA)
Mobile phase: MeOH:buffer 42:58 (Buffer was 5 mM tetraethylammonium acetate adjusted to pH 3.6 with glacial acetic acid.)
Column temperature: 30
Flow rate: 0.8
Injection volume: 10
Detector: UV 230

CHROMATOGRAM
Retention time: 34

OTHER SUBSTANCES
Simultaneous: 7-ACA, 7-ADCA, cefaclor, cefaloridine, cefazolin, cefoperazone, ceftazidime, cephalosporin C

REFERENCE
Tsou,T.-L.; Wu,J.-R.; Wang,T.-M. The effects of separation of cephalosporins by HPLC with β-cyclodextrin bonded stationary phase, *J.Liq.Chromatogr.Rel.Technol.*, **1996**, *19*, 1081–1095.